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# INFLUENCE OF MICROWAVE IRRADIATION ON KINETICS AND THERMODYNAMICS OF EXTRACTION OF FLAVONOIDS FROM *Phyllanthus emblica*

R. Yedhu Krishnan and K.S. Rajan*

Centre for Nanotechnology & Advanced Biomaterials (CeNTAB)
School of Chemical & Biotechnology, SASTRA University, Thanjavur – 613401, India.
*Email: ksrajan@chem.sastra.edu; Phone: +91 9790377951; Fax: +91 4362 264120

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**Abstract –** The effects of solvent-to-feed (S) and temperature (T) on the kinetics and thermodynamics of the microwave assisted extraction (MAE) of flavonoids from dried fruits of *Phyllanthus emblica* using water as solvent have been studied. The flavonoid concentration – time data was analyzed using second order and diffusion models. Both the temperature and solvent-to-feed ratio influenced the initial extraction rate and equilibrium concentration. Biot numbers were found to be greater than 50, implying that the internal mass transfer was the rate controlling step in MAE. The composition of extracts obtained using MAE and conventional extraction was similar. However, the effective diffusion coefficients were doubled in MAE when compared to conventional extraction. An empirical correlation was developed to predict the kinetics of MAE as a function of solvent-to-feed ratio and temperature. The study will be of interest to food and pharmaceutical industries.

*Keywords*: Microwave assisted extraction, *Phyllanthus emblica*, flavonoids, kinetics, thermodynamics

## INTRODUCTION

Flavonoids and other polyphenols are effective antioxidants and free radical scavengers (Garcia-Castello et al., 2015), thereby evincing immense interest among the scientific community. These are secondary plant metabolites that stimulate toxicological or pharmacological effects when consumed (Bernhoft, 2010). In plants, their role in gene regulation and growth metabolism is well established. The average daily intake of flavonoids in the normal diet by humans is about 1 – 2 g (Havsteen, 2002).

*Phyllanthus emblica* (*P. emblica* ) is a euphorbiaceous plant available in Asia (Liu et al., 2008a). The fruits of *P. emblica* are highly nutritive, being rich sources of vitamin 'C' and superoxide dismutase with extensive medicinal uses. The fruit is also a rich source of phenolics (Kusirisin et al., 2009). *P. emblica* fruits are used as laxative, acidulant and diuretic (Gowri et al., 2001). *P. emblica* fruits possess therapeutic activity against diarrhoea, dysentery, haemorrhage, anaemia and dyspepsia (Gowri et al., 2001). The fruits of *P. emblica* are highly perishable and therefore, dehydration of fruits for preservation is essential. Even though several methods are available for dehydration of fruits, open sun-drying is the most widely practiced method (Verma and Gupta, 2004). Dried fruits of *P. emblica* are one of the constituents of the Ayurvedic medicinal preparation *Triphala*, the other being fruits of *Terminalia chebula* and *Terminalia bellerica* (Krishnamachary et al., 2012).

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* To whom correspondence should be addressed

Traditional methods such as soxhlet and conventional heat-reflux extraction for the recovery of bioactive compounds are laborious (Paviani et al., 2012). They require heavy capital investments, longer extraction time, higher solvent consumption and, therefore, are less friendly to the environment (Barba et al., 2015; Dong et al., 2014; Li et al., 2013; Liao et al., 2015; Wakte et al., 2011). However, these shortcomings can be addressed using a non-ionizing electromagnetic radiation – microwave, that can accelerate extraction (Dahmoune et al., 2015; Pavlović et al., 2013). The microwave heats the liquid present in the solid rapidly and partitions the bioactive compounds between solid and solvent phases (Pérez-Serradilla and Luque de Castro, 2011). Water extract of dried fruits of *P. emblica* is used in the preparation of various Ayurvedic formulations, an example being *Lauha bhasma* – a herbo-metallic preparation (Krishnamachary et al., 2013). There has been no report on the application of microwaves for the extraction of flavonoids from fruits of *P. emblica*. Therefore, a study on microwave assisted extraction (MAE) of active compounds from sun-dried fruits of *P. emblica* using water as the solvent assumes importance.

The kinetics of microwave-assisted water extraction of flavonoids from *P. emblica* has not been reported in the literature. It is pertinent to note that the knowledge of the kinetics is essential for the design of extraction systems (Baümler et al., 2014; Honarvar and Sajadian, 2013; Minozzo et al., 2012)

In the present work, the influence of temperature (T) and solvent-to-feed ratio (S) on the kinetics and thermodynamics of microwave-assisted extraction of flavonoids from *P. emblica* is reported. It is pertinent to mention that no prior art on the kinetics and thermodynamics of MAE of flavonoids from *P. emblica* is available in the literature.

## MATERIALS AND METHODS

### Plant material

Commercially available dried fruits of *P. emblica* authenticated by CARISM, SASTRA University, Thanjavur, India were used for the experiments. The seeds were removed from the dried fruits. The powder was screened in a rotary sieve shaker and particles that passed through BSS 200 but were retained on BSS 300 (average particle size ~ 0.064 mm) were selected. Fine particles were preferred to achieve higher interfacial area for mass transfer. The particle size (64 μm) was chosen to lie closer to the average size of plant cells.

### Chemicals & reagents

The reagents required for estimation of total flavonoid content such as aluminium nitrate and potassium acetate were procured from Hi Media Ltd., Mumbai, India. Quercetin, used as the standard for calibration, was procured from Aldrich Chemical Company, USA. MAE was carried out using distilled water as the solvent.

### Microwave-assisted extraction (MAE)

A 22-litre, 800 W microwave oven (R-219T (S)/(W), SHARP, Japan) was employed for the experiments. 5 g of dried fruits of *P. emblica* was measured into a round-bottomed flask with varying amount of water (solvent), ranging from 25 mL to 200 mL and subjected to microwave irradiation. A reflux condenser was mounted onto the flask as shown in the schematic diagram (Figure 1). The reflux condenser was cooled continuously using cold water to prevent solvent loss by vaporization. Ideal mixing conditions were simulated using a magnetic stirrer placed beneath the flask. A digital temperature indicator was used for temperature measurement. The desired temperature during MAE was maintained through the timer. Water was used as the solvent for the extraction of flavonoids from the dried fruits of *P. emblica*. A small volume of sample (250 μL) was removed at different time points.

### Conventional extraction (CE)

For the purpose of determination of total flavonoid concentration in the solid matrix, a 3-stage conventional heat-reflux extraction of *P. emblica* with different solvents was performed. Extraction was first carried out with chloroform as the solvent, followed by methanol and water as solvents for the second and third stages. The duration of each stage of extraction was 12 hours. A very high solvent-to-feed ratio (50 mL/g) was used during extraction with each solvent to ensure complete recovery of flavonoids, for the purpose of determination of total flavonoid concentration. In addition, the conventional heat-reflux extraction of flavonoids from *P. emblica* using water was carried out for comparison with MAE.

### Experimental design

Experiments were performed according to full factorial design using four levels each for the solvent-to-feed ratio and temperature. The salient physical properties of water used as the solvent for extraction are shown in Table 1. A total of 48 experimental trials (16 experimental conditions in triplicates) were performed with MAE. All results are reported as mean ± standard error of the mean. Table 2 shows the experimental design with actual values of levels in parentheses and the corresponding values of the equilibrium concentration of flavonoids ($C_s$).

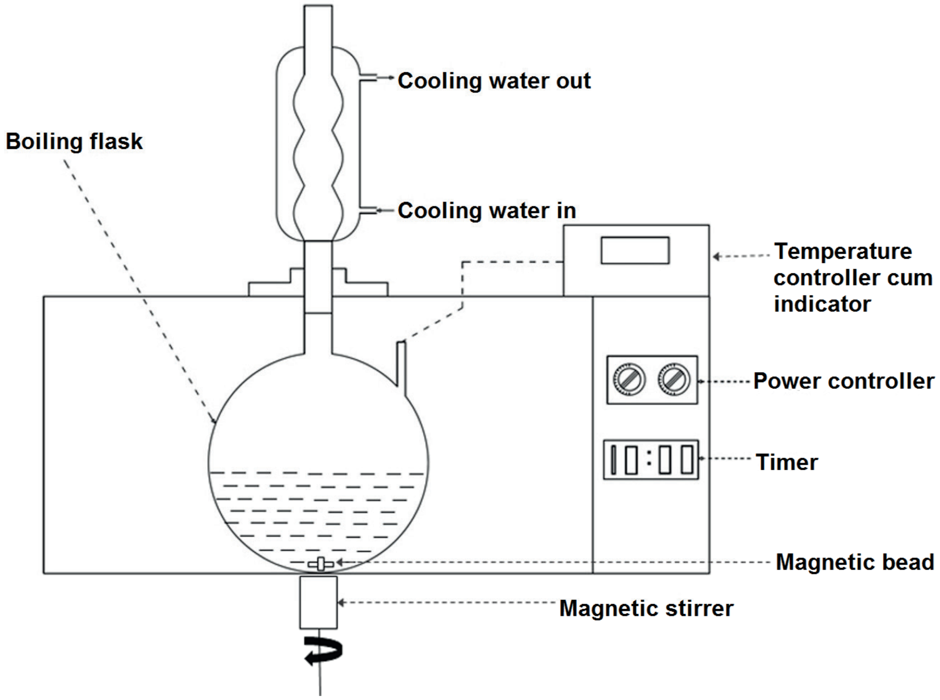

**Figure 1.** Experimental set for microwave-assisted extraction.

**Table 1.** Physical properties of water used for the extraction.

| Property | Value |
|---|---|
| Electrical conductivity | 6.8 µS/m |
| Thermal conductivity | 0.6 W/(m K) |
| Density | 1005 kg/m$^3$ |
| Viscosity | 7.8×10$^{-4}$ N s/m$^2$ |

**Table 2.** Details of the full factorial experimental design.

| Run Numbers | A: Solvent-to-feed ratio Level (Value in mL/g) | B: Temperature Level (Value in °C) | $C_s$ (mgQE/mL) |
|---|---|---|---|
| 1, 28, 34 | 4 (40) | 4 (100) | 0.38±0.01 |
| 2, 7, 42 | 4 (40) | 3 (80) | 0.34±0.00 |
| 3, 19, 43 | 4 (40) | 1 (40) | 0.28±0.00 |
| 4, 33, 48 | 1 (5) | 2 (60) | 0.58±0.00 |
| 5, 11, 15 | 2 (10) | 1 (40) | 0.41±0.00 |
| 6, 18, 24 | 3 (20) | 1 (40) | 0.37±0.01 |
| 8, 30, 38 | 3 (20) | 4 (100) | 0.54±0.00 |
| 9, 21, 32 | 1 (5) | 3 (80) | 0.64±0.00 |
| 10, 22, 47 | 1 (5) | 1 (40) | 0.52±0.00 |
| 12, 31, 37 | 2 (10) | 3 (80) | 0.54±0.02 |
| 13, 35, 44 | 2 (10) | 4 (100) | 0.67±0.01 |
| 14, 23, 27 | 1 (5) | 4 (100) | 0.79±0.00 |
| 16, 20, 25 | 4 (40) | 2 (60) | 0.32±0.00 |
| 17, 39, 40 | 3 (20) | 3 (80) | 0.47±0.01 |
| 26, 29, 45 | 3 (20) | 2 (60) | 0.41±0.01 |
| 36, 41, 46 | 2 (10) | 2 (60) | 0.47±0.01 |

### Analytical method

Aluminium nitrate assay (Sen et al., 2013; Shah and Hossain, 2014) was used for the determination of total flavonoid content. 500 µL of the test sample, 100 µL of aluminium nitrate (10 %), 100 µL of potassium acetate (1 M) and 4.3 mL of distilled water were mixed well and incubated at room temperature for 40 min. The absorbance of the mixture was measured at 415 nm (Lambda 25, Perkin Elmer, USA) against a reagent blank comprising aluminium nitrate, potassium acetate and water alone. The standard curve was prepared using Quercetin as the standard. The flavonoid concentrations are expressed as mg quercetin equivalent per mL of solvent (mgQE/mL).

### Gas chromatography – mass spectrometry (GC-MS)

GC-MS analysis of representative dried and re-constituted extracts was carried out using a gas chromatograph (PerkinElmer Clarus 500) equipped with a capillary column (30 m × 0.25 mm). About 2 µL of the sample was injected using helium as the carrier gas supplied at 1 mL/min. The temperature of the oven was programmed to ramp from 60 ºC to 150 ºC at the rate of 6 ºC/min and maintained at 150 ºC for 2 minutes. This was followed by further heating at the rate of 4 ºC/min to achieve the desired injector temperature of 280 ºC, and was maintained for 5 minutes. A scan range of 40 - 450 amu was employed during the recording of mass spectra in electron ionization mode. The NIST 2005 Mass spectral library was used for the identification of the constituents of the extracts.

### Uncertainty

Uncertainty in the measurement of temperature, mass of the feed and flavonoid concentration is shown in Table 3.

**Table 3.** Uncertainty in the measurements.

| Measurement | Uncertainity |
|---|---|
| Temperature | ±1 °C |
| Mass of feed | ±1 mg |
| Flavonoid concentration | ±10 µg/mL |

## RESULTS AND DISCUSSION

### Modelling of kinetics of MAE

The kinetics of extraction may be explained using a second-order model (Ho et al., 2005; Qu et al., 2010) as follows:

$$\frac{dC_t}{dt} = k(C_s - C_t)^2 \tag{1}$$

In Eq. (1), $C_t$ is the concentration of flavonoids (mgQE/mL) at any time $t$, $C_s$ is the equilibrium concentration of flavonoids (mgQE/mL) and $k$ is the second order rate constant (mL/mgQE.min). The solution of Eq. (1) with appropriate boundary conditions is:

$$C_t = \frac{C_s^{\,2} kt}{1 + C_s kt} \tag{2}$$

Rearranging Eq. (2), we get

$$\frac{t}{C_t} = \frac{1}{kC_s^{\,2}} + \frac{t}{C_s} \qquad (3$$

The product of 'k' and '$C_s^{\,2}$' is the initial rate 'h'. Accordingly Eq. (3) becomes

$$\frac{t}{C_t} = \frac{1}{h} + \frac{t}{C_s} \tag{4}$$

The equilibrium concentration $C_s$ (mgQE/mL), extraction rate constant $k$ (mL/mgQE.min) and initial extraction rate $h$ (mgQE/mL.min) can be determined from a graph of $t/C_t$ vs $t$.

### Estimation of effective diffusion coefficient, mass transfer coefficient and Biot number

Fick's second law was used to model diffusion during the MAE of flavonoids from *P. emblica*, assuming particles to be spherical. Mass transfer resistance in the liquid phase was considered to be negligible due to continuous stirring during MAE. No degradation of flavonoids was expected during MAE under the present experimental conditions, as flavonoid degradation is reported to begin at a temperature greater than 110 ºC (Xiao et al., 2008).

The change in flavonoid concentration in the solid phase with respect to time and radial position during MAE can be described using Fick's second law as follows (Tao et al., 2014a):

$$\frac{\partial M}{\partial t} = D_e\left[\frac{1}{r^2}\frac{\partial}{\partial r}\left(r^2\frac{\partial M}{\partial r}\right)\right] \tag{5}$$

In Eq. (5), $M$ is the flavonoid concentration in the *P. emblica* fruit particle (kg/m$^3$), $r$ is the distance from the centre (m).

The solution of Eq. (5) using the appropriate initial and boundary conditions is (Schwartzberg, 1975):

$$\frac{Y_t}{Y_s} = 1 - \frac{6}{\pi^2}\exp\left(-\frac{D_e \pi^2 t}{Rp^2}\right) \tag{6}$$

In Eq. (6), $Y_t$ is the total flavonoid (mgQE/g) transferred (Yield) from *P. emblica* fruit at time *t*, $D_e$ is the effective diffusion coefficient and $Y_s$ is the total flavonoid transferred (mgQE/g) at equilibrium. The effective diffusion coefficient ($D_e$) can be estimated from the experimental data using the linearized form of Eq. (6).

The Biot number (Bi) provides an estimation of the relative magnitudes of external (film resistance) and internal resistances (diffusion) to mass transfer. The Biot number is related to particle size ($D_p$) mass transfer coefficient ($K_T$) and effective diffusion coefficient ($D_e$) as (Rakotondramasy-Rabesiaka et al., 2010):

$$Bi = \frac{D_p K_T}{D_e} \tag{7}$$

The rate of mass transfer of solute from the solid phase to the liquid phase is related to the liquid phase volume and rate of change of solute concentration in the liquid as (Berthier, 1952):

$$V_L \frac{dC_t}{dt} = AK_T\left(C_s - C_t\right) \tag{8}$$

where A is the surface area ($m^2$) and $V_L$ is the volume of solvent ($m^3$).

Eq. (8) can be integrated using appropriate initial and boundary conditions as follows:

$$\mathrm{h}\ \frac{C_s}{C_s - C_t} = \frac{K_T A}{V_L} t \tag{9}$$

Eq. (9) is valid till $t \leq t_R$, where $t_R$ is the regular regime starting time, which can be estimated from the second order kinetic model by substituting $C_t$ as $\frac{C_s}{2}$ at t = $t_R$ (Rakotondramasy-Rabesiaka et al., 2010). Therefore, the expression for '$t_R$' relating 'k' and '$C_s$' is given as follows (Rakotondramasy-Rabesiaka et al., 2010):

$$t_R = \frac{1}{kC_s} \tag{10}$$

## Estimation of thermodynamic parameters of MAE

Thermodynamic analysis of the extraction of solute from the solid by MAE can be carried out through determination of changes in enthalpy (ΔH°), entropy (ΔS°) and Gibbs free energy (ΔG°).

The equilibrium constant for leaching ($K_e$) is defined as the ratio of the amount of solute removed from the solid to the amount of solute remaining in the solid (Meziane and Kadi, 2008). The total flavonoid available in the solid matrix ($Y_{max}$) determined using exhaustive extraction with three solvents was found to be 21.69 mgQE/g. This value closely matches with the flavonoid content in *P. emblica* reported earlier (Liu et al., 2008b). Therefore, 'Ke' is the ratio of flavonoid yield at equilibrium ($Y_s$) to the amount of flavonoid remaining in the solid at equilibrium ($Y_{max}$-$Y_S$) as given below:

$$K_e = \frac{Y_s}{(Y_{\max} - Y_s)} \tag{11}$$

ΔH° and ΔS° are related to '$K_e$', the universal gas constant (R) and temperature (T) using the Van't Hoff equation (Meziane and Kadi, 2008) as follows:

$$\ln K_e = -\frac{\Delta H^\circ}{RT} + \frac{\Delta S^\circ}{R} \tag{12}$$

A plot of ln $K_e$ vs 1/T can be used determine ΔH° and ΔS° from the slope and intercept, respectively.

The change in Gibbs free energy (ΔG°) is related to ΔH°, temperarature and ΔS° as follows (Meziane and Kadi, 2008):

$$\Delta G^\circ = \Delta H^\circ - T\Delta S^\circ \tag{13}$$

The change in entropy (ΔS°) is a degree of randomness or the measure of unavailable energy in a closed thermodynamic system. A positive value of the change in entropy (ΔS°) implies an increase in disorder of the system (Amarante et al., 2014). A negative value for the change in Gibbs free energy (ΔG°) implies spontaneity of the process. Endothermic processes are associated with a positive value of the change in enthalpy (ΔH°).

## Suitability of second-order and diffusion models for extraction kinetics

The concentration of total flavonoids extracted at different periods of extraction is shown in Figure 2 at a solvent-to-feed ratio of 40 mL/g. It can be noted from

Figure 2 that there are two phases in the release kinetics. During the initial phase of extraction, there is a swift increase in the concentration due to rapid dissolution of the solute from the particle surface through a washing mechanism. This is followed by a more gradual diffusion phase (Meziane and Kadi, 2008). Therefore, the second-order model can be utilized to explain the non-linear total flavonoid concentration–time relationship.

The kinetic parameters (h, k) and $C_s$ of the second order kinetic model in the temperature range of 40 – 100 °C and the solvent-to-feed ratio of 40 mL/g for MAE are shown in Table 4. The values of h, k and $C_s$ for CE under the same conditions of temperature and solvent-to-feed ratio are shown in Table 5. The second order model describes the kinetics of microwave assisted extraction satisfactorily at all experimental conditions (Figure 3) with an average absolute relative deviation of 0.802%, relative standard deviation of 1.491% and $R^2$ of 0.997. A comparison of Tables 4 and 5 reveals that the initial extraction rate and extraction rate constant increase upon microwave irradiation.

The suitability of Fick's equation for the fitness of kinetics of MAE is evident from the closeness of the predicted and experimental total flavonoid yield data for t≥0.5 min (Figure 4), with the average absolute relative deviation, relative standard deviation and $R^2$ being 1.691%,

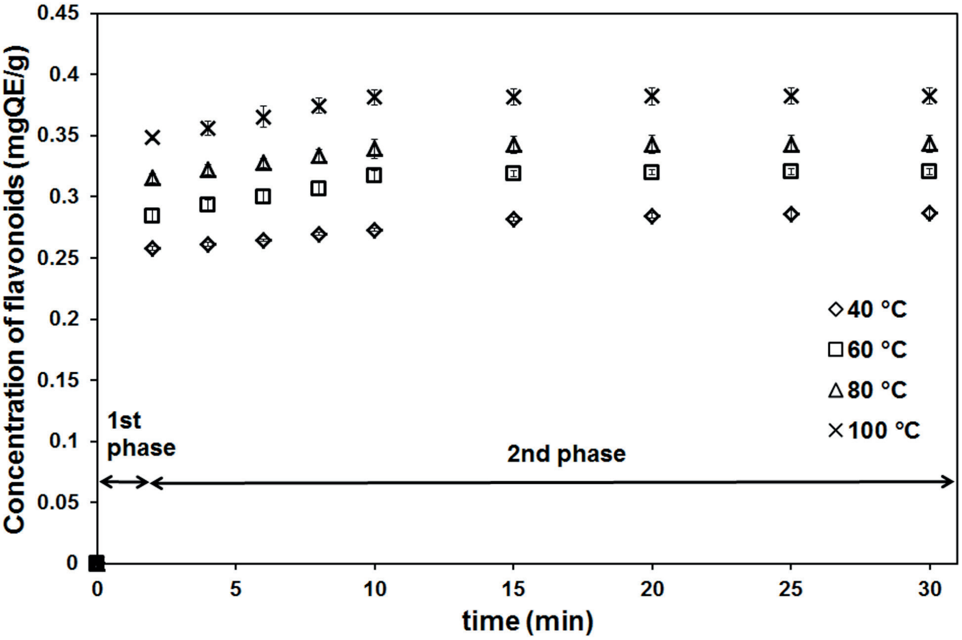

**Figure 2.** Influence of extraction time on the concentration of flavonoids during MAE carried out at the solvent-to-feed ratio of 40 mL/g.

**Table 4.** Kinetic parameters (h, k) and $C_s$ of the second order model for MAE at the solvent-to-feed ratio of 40 mL/g.

| T (°C) | $C_s$ (mgQE/mL) | h (mgQE/mL.min) | k (mL/mgQE.min) | $R^2$ |
|---|---|---|---|---|
| 40 | 0.28±0.00 | 0.81±0.03 | 9.98±0.47 | 0.99 |
| 60 | 0.32±0.00 | 1.29±0.10 | 12.48±0.82 | 0.99 |
| 80 | 0.34±0.00 | 1.76±0.05 | 14.56±0.20 | 0.99 |
| 100 | 0.38±0.01 | 2.34±0.03 | 15.89±0.41 | 0.99 |

**Table 5.** Kinetic parameters (h, k) and $C_s$ of the second order model for CE at the solvent-to-feed ratio of 40 mL/g.

| T (°C) | $C_s$ (mgQE/mL) | h (mgQE/mL.min) | k (mL/mgQE.min) | $R^2$ |
|---|---|---|---|---|
| 40 | 0.28±0.00 | 0.16±0.05 | 2.08±0.13 | 0.99 |
| 60 | 0.30±0.00 | 0.21±0.05 | 2.35±0.04 | 0.99 |
| 80 | 0.33±0.00 | 0.32±0.07 | 2.85±0.08 | 0.99 |
| 100 | 0.35±0.00 | 0.42±0.09 | 3.29±0.20 | 0.99 |

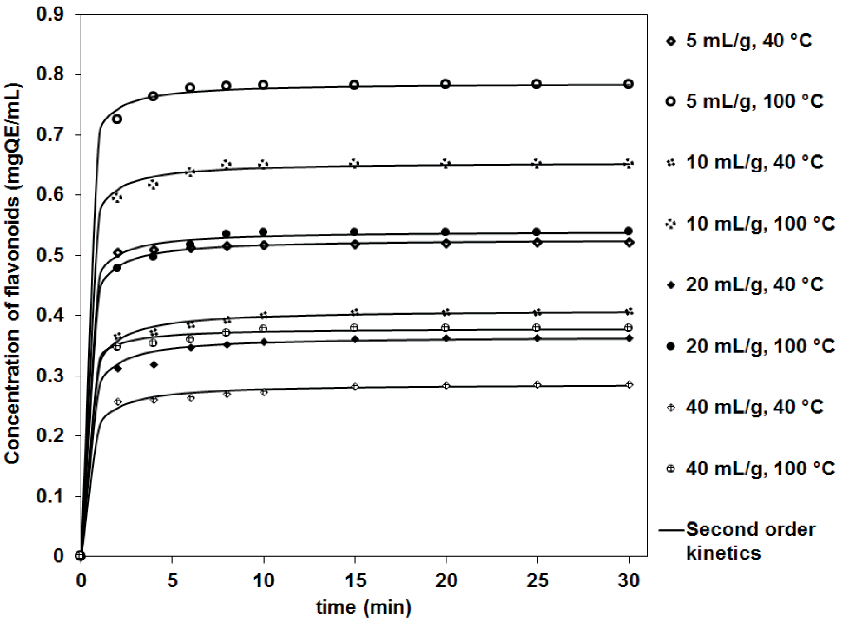

**Figure 3.** Application of the second order model for the kinetics of MAE.

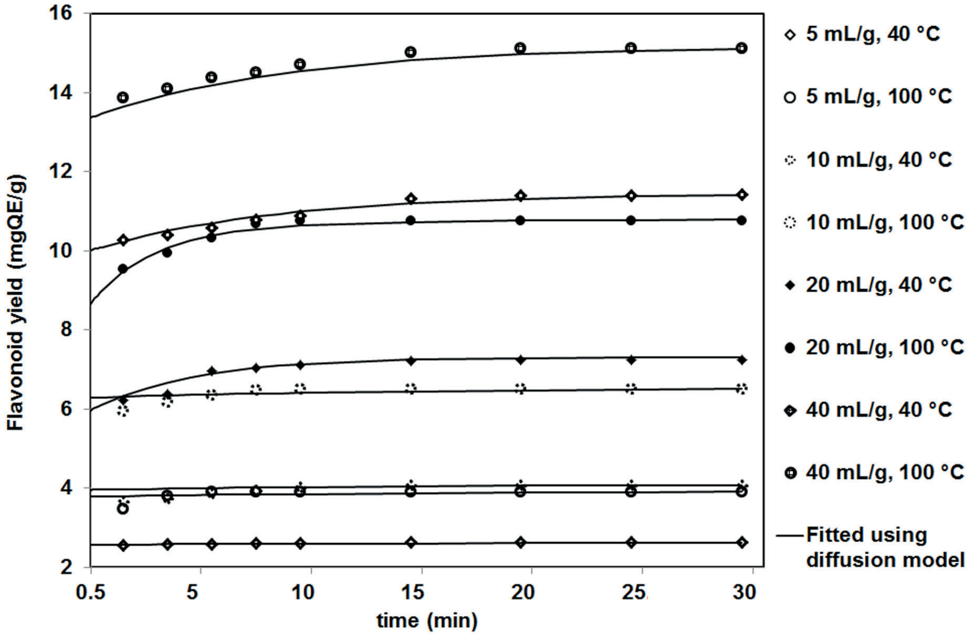

**Figure 4.** Suitability of the diffusion model for MAE of flavonoids from *P. emblica*.

4.057% and 0.997 respectively. Equation 6 does not apply as tà0 and hence predictions have been made starting with extraction time of 30 seconds (0.5 min).

## Effect of solvent-to-feed ratio

The effect of solvent-to-feed ratio on the equilibrium concentration of flavonoids, equilibrium yield and initial extraction rate is shown in Figures 5 – 7, respectively, at 40 °C and 100 °C. At higher solvent-to-feed ratios, the equilibrium solute concentration in the extract was reduced (Figure 5). This might be attributed to the larger volume of solvent used at higher solvent-to-feed ratios. It is clear from Figure 6 that, with the increase in solvent-to-feed ratio, the equilibrium yield of flavonoids increased. During extraction, the interphase mass transfer occurs in both directions. The concentration gradient for solute drives the mass transfer from solid to the liquid phase

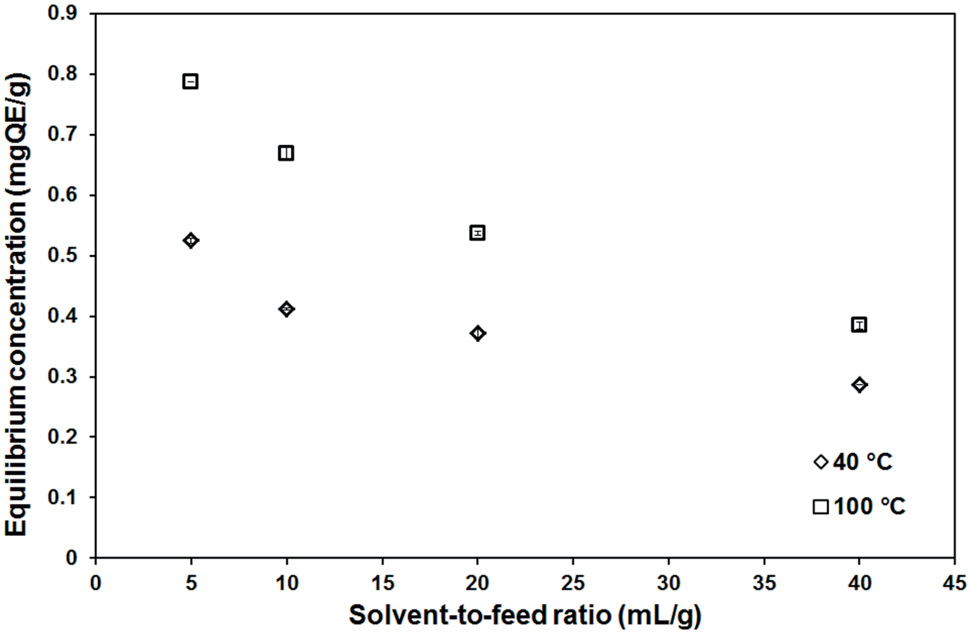

**Figure 5.** Influence of solvent-to-feed ratio on the equilibrium concentration at T=40 °C and T= 100 °C for MAE.

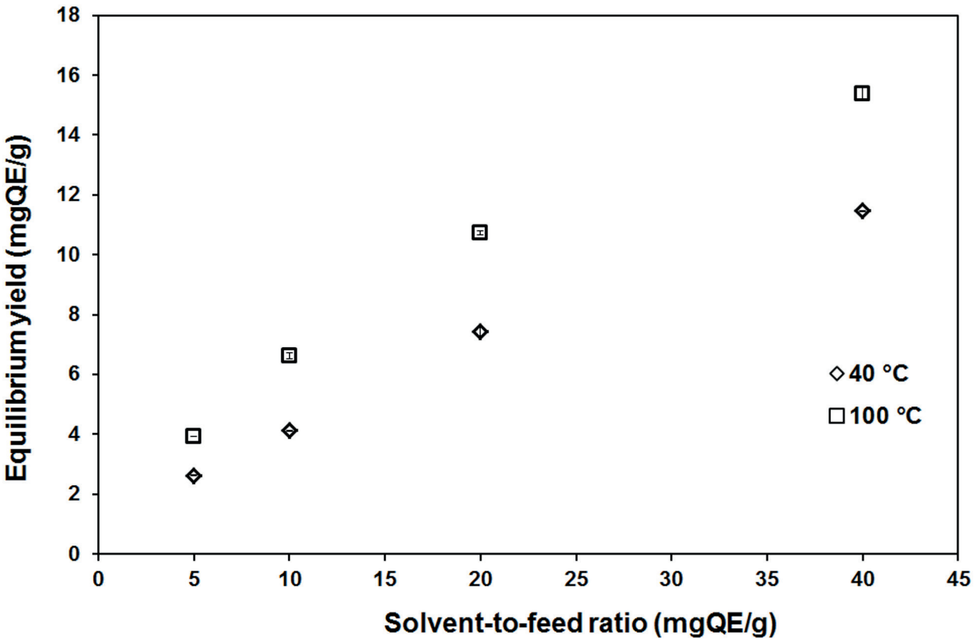

**Figure 6.** Influence of solvent-to-feed ratio on the equilibrium yield at T=40 °C and T=100 °C for MAE.

by dissolution and diffusion. However, solute can also adsorb non-preferentially on the solid surface as well. At equilibrium, the solute concentration in the liquid phase at higher solvent-to-feed ratios was lower (Figure 5). This resulted in an increase in the driving force for dissolution and diffusion. In addition, lower solute concentration reduced the driving force for adsorption. Hence, higher equilibrium yields were obtained at higher solvent-to-feed ratio. The effect of solvent-to-feed ratio on initial extraction rate is shown in Figure 7, from which it is clear that the initial extraction rate was higher at the lower solvent-to-feed ratio. The higher initial rate at the lower solvent-to-feed ratio might be attributed to the fact that the initial extraction is dominated by the washing phase, where the solvent washes out the solute rapidly.

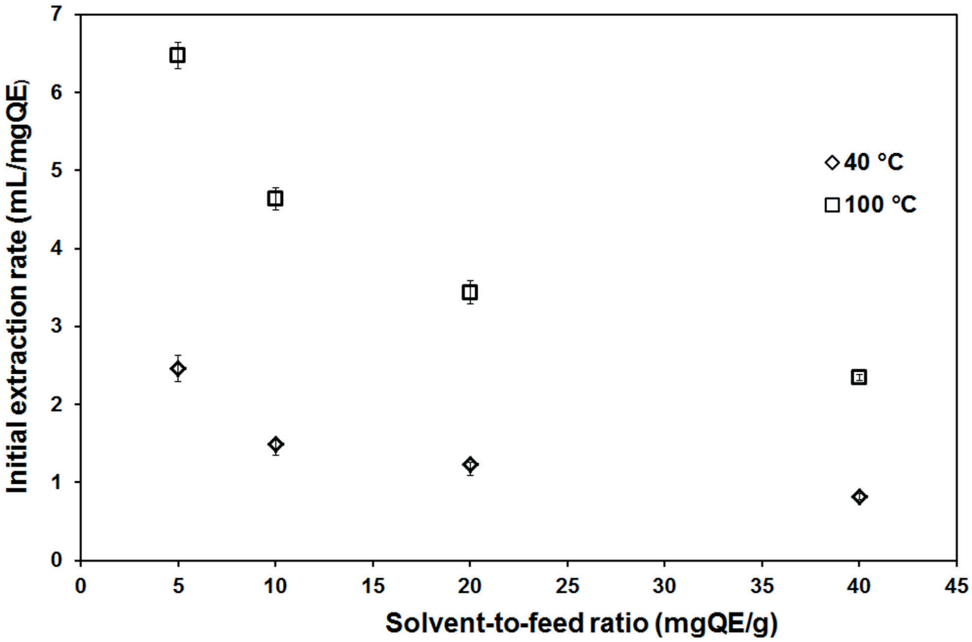

**Figure 7.** Effect of solvent-to-feed ratio on initial extraction rate at T=40 °C and T=100 °C for MAE.

## Effect of temperature

The effect of temperature (T) on the equilibrium concentration and initial extraction rate for MAE is shown in Figures 8 & 9 respectively. With an increase in temperature, the equilibrium concentration increased at all solvent-to-feed ratios investigated (Figure 8). With an increase in temperature, the solute-solute and solute-solid interactions are weakened, leading to the reduced affinity of the solid for the solute. Also higher temperatures aid in desorption of the solutes from active sites within the plant matrix (Veggi et al., 2013). The solute transfer from the solid-to-liquid phase is affected by transport properties such as effective diffusion coefficient (Tao et al., 2014a). With the increase in temperature, the effective diffusion coefficient for MAE of flavonoids also increased (Table 6) due to higher thermal energy at elevated temperatures. Similarly, the effective difusion coefficient for CE of flavonoids from *P. emblica* is shown in Table 6. The effective diffusion coefficients in MAE were found to be higher than those in CE, indicating the ease of solute transfer during both the washing and diffusion steps in MAE.

The increased mass transfer coefficient might have assisted in increasing the initial extraction rate (Figure 9). The higher values of Biot number (>50) indicate that the external resistance (film resistance) for mass transfer was insignificant due to turbulence at the solid-liquid interface caused by mixing (Tao et al., 2014b). Therefore, internal mass transfer (diffusion) is rate-limiting (Rakotondramasy-Rabesiaka et al., 2010).

The activation energy ($E_a$) for MAE is related to the extraction rate constant (k) and the temperature (T) as follows:

$$k = k_0 . e^{\frac{-E_a}{RT}} \tag{14}$$

The activation energy, which is an energy barrier for solute transfer, can be determined by linearizing Eq. (14) as shown in Figure 10.

The activation energy, arising mainly from solute-solute cohesion and solute-solid adhesion (Alupului et al., 2012) was estimated as 7.58 kJ/mol. The microwave-assisted extraction process is endothermic and spontaneous, as evident from the positive and negative values for ΔH° and ΔG° respectively (Table 8). The increase in entropy due to extraction is attributed to solute transfer from the solid phase to the liquid phase (Amarante et al., 2014). With an increase in temperature, the degree of spontaneity of the MAE increases, leading to favourable extraction at elevated temperatures. A comparison of thermodynamic parameters of MAE (Table 8) with those of CE (Table 9) reveals that MAE is more spontaneous than CE.

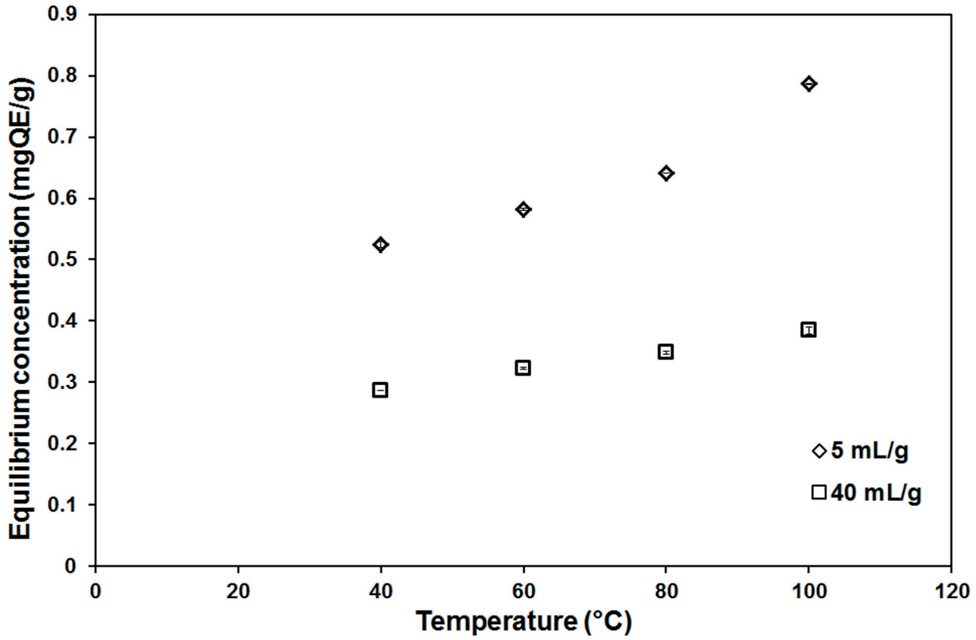

**Figure 8.** Effect of temperature on the equilibrium concentration at S=5 mL/g and S=40 mL/g for MAE.

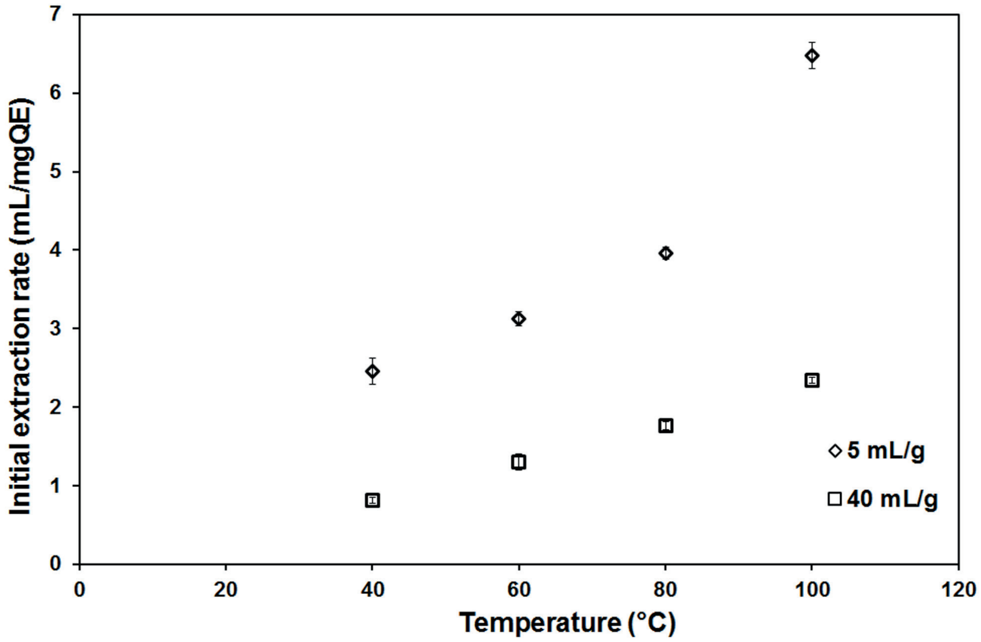

**Figure 9.** Effect of temperature on the initial extraction rate at S=5 mL/g and S=40 mL/g for MAE.

**Table 6.** Effective diffusion coefficient for MAE and CE of flavonoids from *P. emblica* at a solvent-to-feed ratio of 40 mL/g.

| T (°C) | MAE | | CE | |
|---|---|---|---|---|
| | $D_e \times 10^{11}$ (m$^2$/s) | $R^2$ | $D_e \times 10^{11}$ (m$^2$/s) | $R^2$ |
| 40 | 1.15±0.02 | 0.98 | 0.63±0.06 | 0.99 |
| 60 | 1.74±0.01 | 0.99 | 0.69±0.03 | 0.97 |
| 80 | 1.95±0.02 | 0.98 | 0.82±0.05 | 0.97 |
| 100 | 2.08±0.10 | 0.98 | 0.86±0.02 | 0.99 |

**Table 7.** Mass transfer coefficient and Biot number for MAE of flavonoids from *P. emblica* at a solvent-to-feed ratio of 40 mL/g.

| T (°C) | $K_T \times 10^5$ (m/s) | Bi |
|---|---|---|
| 40 | 1.26±0.23 | 73.26±2.28 |
| 60 | 2.21±0.00 | 82.06±1.23 |
| 80 | 2.74±0.00 | 92.14±0.44 |
| 100 | 3.24±0.10 | 106.56±3.59 |

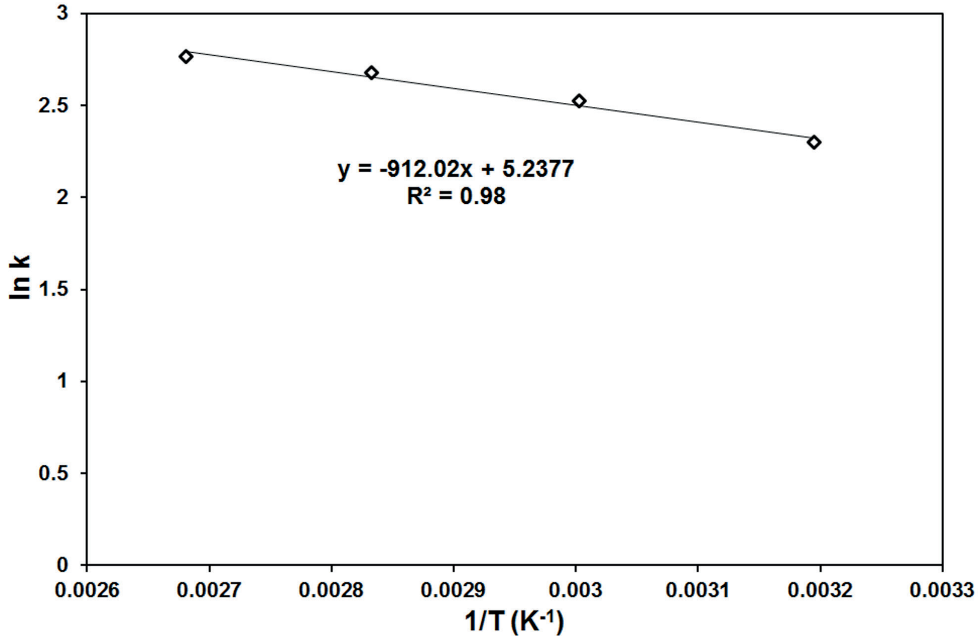

**Figure 10.** Determination of the activation energy for extraction.

**Table 8.** Thermodynamic parameters for MAE of flavonoids from *P. emblica.*

| T (K) | $K_e$ | ΔH° (kJ/mol) | ΔS° (J/mol.K) | ΔG° (kJ/mol) |
|---|---|---|---|---|
| 313 | 1.11 | 12.25 | 39.96 | -0.25 |
| 333 | 1.46 | | | -1.05 |
| 353 | 1.79 | | | -1.85 |
| 373 | 2.43 | | | -2.65 |

**Table 9.** Thermodynamic parameters for CE of flavonoids from *P. emblica.*

| T (K) | $K_e$ | ΔH° (kJ/mol) | ΔS° (J/mol.K) | ΔG° (kJ/mol) |
|---|---|---|---|---|
| 313 | 1.09 | 9.53 | 30.93 | -0.15 |
| 333 | 1.26 | | | -0.76 |
| 353 | 1.59 | | | -1.38 |
| 373 | 1.94 | | | -2.00 |

## Development of empirical correlations

The experimental data for the dependence of temperature and solvent-to-feed ratio on 'h' and '$C_s$' were analyzed statistically. The empirical correlations for the dependence of equilibrium flavonoid concentration ($C_s$) and initial extraction rate (h) on solvent-to-feed ratio and temperature, valid for $40 \le T \le 100°C$ and $5 \le S \le 40 mL/g$, are as follows:

$$h = 0.0620(T)^{1.16}(S)^{-0.454} \tag{15}$$

$$C_s = 0.1845(T)^{0.409}(S)^{-0.297} \tag{16}$$

Substituting Eqs. (15) and (16) in Eq. (4), a $C_t$-t-T-S relationship is obtained as follows (valid for $40 \le T \le 100°C$ and $5 \le S \le 40 mL/g$ ):

$$C_t = \frac{t}{\dfrac{1}{0.0620(T)^{1.16}(S)^{-0.454}} + \dfrac{t}{0.1845(T)^{0.409}(S)^{-0.297}}} \tag{17}$$

The inspection of Eqs. (15) and (16) proves that the temperature has a higher influence on 'h' and '$C_s$' than that of solvent-to-feed ratio, as evident from the higher absolute values of the exponents.

The applicability of Eq. (17) for prediction of extraction kinetics was tested using the experimental data acquired under four new sets of experimental conditions, viz., T=50 °C, S=40 mL/g; T=80 °C, S=30 mL/g; T=60 °C, S=15 mL/g and T=70 °C, S=10 mL/g. Figure 11 shows the applicability of empirical correlations to the new experimental kinetic data that were not used in the development of the empirical correlations. Eq. (17) predicted the kinetic data with an average absolute relative deviation of 2.205 %, relative standard deviation of 4.042 % and $R^2$ of 0.971.

## Chemical compositions of extracts of *P. emblica* fruits

The composition of the microwave-assisted aqueous extract of *P. emblica* fruits prepared at 40 °C and 100 °C using the solvent-to-feed ratio of 40 mL/g is given in Table 10. The composition of the extract prepared by conventional extraction under identical conditions is also provided in Table 10. The major flavonoid in the extract is, 2,3-dihydro-3,5-dihydroxy-6-methyl-4H-Pyran-4-one, present to the extent of 1.0402% by MAE at 100 °C . All the major compounds present in the microwave-assisted extract of *P. emblica* fruits were observed to be identical to those isolated by a conventional extraction process. In addition, the composition of extracts prepared at 40 °C and 100 °C is similar, indicating that the flavonoids were not degraded even at 100 °C.

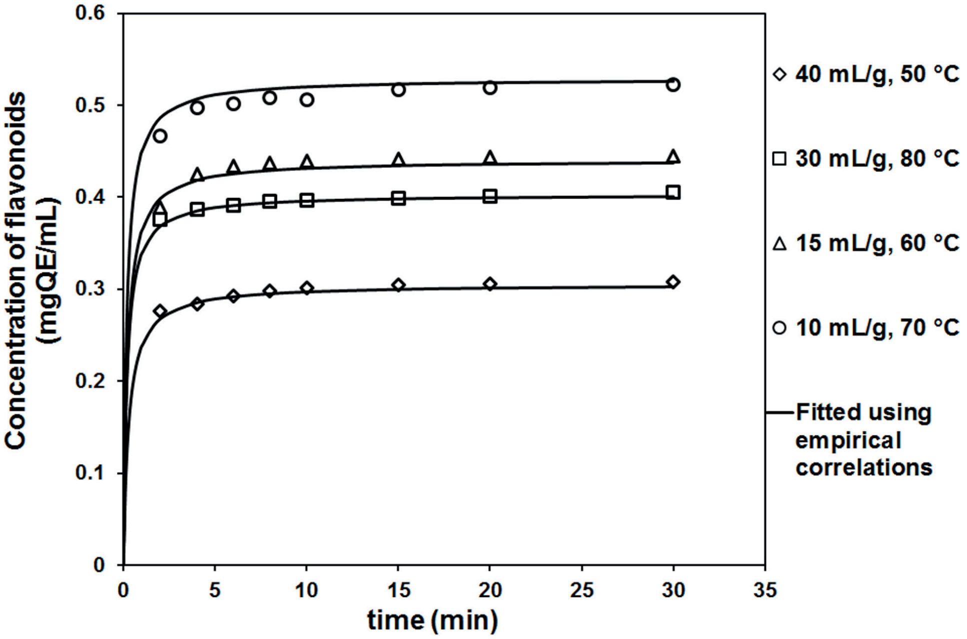

**Figure 11.** Applicability of Eq. (17) in predicting the new kinetic data.

**Table 10.** Composition of the aqueous extract of *P. emblica* fruits at T=40 °C and T=100 °C.

| Compounds | % Peak area | | | |
|---|---|---|---|---|
| | MAE | | CE | |
| | 40 °C | 100 °C | 40 °C | 100 °C |
| 4H-Pyran-4-one, 2,3-dihydro-3,5-dihydroxy-6-methyl- | 0.68 | 1.04 | 0.63 | 1.02 |
| 2-Furancarboxaldehyde, 5-(hydroxymethyl)- | 4.28 | 4.46 | 6.11 | 8.37 |
| Benzene, 1,1'-(1-methylethylidene)bis[4-methoxy- | 4.38 | 10.21 | 2.81 | 6.70 |
| 1,2,3-Benzenetriol | 63.27 | 51.57 | 62.24 | 57.47 |
| D-Allose | 7.45 | 21.89 | 10.19 | 12.94 |
| 1,6-Anhydro-α-d-galactofuranose | 1.81 | 1.58 | 1.91 | 3.20 |
| Other minor compounds | 18.09 | 9.22 | 16.08 | 10.26 |

## CONCLUSION

The present work investigated the MAE of flavonoids from *P. emblica* fruits using water as the solvent. The second-order kinetic model and the diffusion model were able to fit the experimental data satisfactorily. The extraction kinetics revealed the existence of two phases – a rapid washing phase and a slower diffusion phase. Both solvent-to-feed ratio and extraction temperature were observed to influence extraction kinetics, with the temperature being the more influential factor. The highest flavonoid yield was obtained at 40 mL/g and 100 °C. Higher solvent-to-feed ratio favoured extraction by producing higher equilibrium yield and higher temperature aided in a higher diffusion coefficient. The study of thermodynamic parameters showed MAE to be spontaneous and endothermic. Overall, this study helped to identify operating conditions for improved flavonoid yield during MAE using water alone as the solvent.

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## NOMENCLATURE

| | |
|---|---|
| A | surface area $m^2$ |
| Bi | Biot number |
| $C_t$ | concentration of flavonoids at time t mgQE $mL^{-1}$ |
| $C_s$ | equilibrium concentration of flavonoids mgQE $mL^{-1}$ |
| $D_e$ | effective diffusion coefficient of flavonoids $m^2\ s^{-1}$ |
| $D_p$ | size of particle m |
| $E_a$ | activation energy for extraction kJ $mol^{-1}$ |
| h | initial extraction rate mgQE $mL^{-1}$ $min^{-1}$ |
| k | extraction rate constant mL $mgQE^{-1}$ $min^{-1}$ |
| $k_0$ | pre-exponential factor mL $mgQE^{-1}$ $min^{-1}$ |
| $K_e$ | equilibrium constant for leaching |
| $K_T$ | mass transfer coefficient m $s^{-1}$ |
| $M_i$ | initial flavonoid concentration in *P. emblica* fruit particle at the start of extraction kg $m^{-3}$ |
| M | flavonoid concentration in *P. emblica* fruit particle kg $m^{-3}$ |
| r | distance from the centre of a spherical *P. emblica* fruit particle m |
| R | gas constant J $mol^{-1}$ $K^{-1}$ |
| Rp | radius of *P. emblica* fruit particle m |
| S | Solvent-to-feed ratio mL $g^{-1}$ |
| $t_R$ | regular regime starting time min |
| T | Temperature K / °C |
| $V_L$ | volume of solvent $m^3$ |
| $Y_t$ | total flavonoids transferred from *P. emblica* fruit at time t mgQE $g^{-1}$ |
| $Y_s$ | total flavonoids transferred from *P. emblica* fruit at equilibrium mgQE $g^{-1}$ |
| $Y_{max}$ | total flavonoids available in *P. emblica* mgQE $g^{-1}$ |
| | ***Greek letters*** |
| $\Delta G°$ | Gibbs free energykJ $mol^{-1}$ |
| $\Delta H°$ | change in enthalpy kJ $mol^{-1}$ |
| $\Delta S°$ | change in entropyJ $mol^{-1}$ $K^{-1}$ |

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